Population Genetics of Mexican *Drosophila*. II. A New Species of the *obscura* Group of the Genus *Drosophila*

(Diptera, Drosophilidae)[1]

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The *obscura* species group of the subgenus *Sophophora* of the genus *Drosophila* consists of 8 Old World and 4 New World species. The latter are *D. pseudoobscura* Frolova, *D. persimilis* Dobzhansky and Epling, *D. miranda* Dobzhansky, and *D. lowei* Heed. They live in the Western United States and Canada; only *D. pseudoobscura* extends its geographic distribution to the highlands of Mexico, Guatemala, and the Andes above Bogotá, Colombia. The related *affinis* species group contains 9 species distributed over North and Central America. We are engaged in studies of the population genetics of species of these groups living in Central Mexico, particularly *D. pseudoobscura*. In the process of collecting population samples for these studies we have discovered a new species to be described in the present article. It clearly belongs to the *obscura* species group, and its females are very similar in external appearance to those of *D. pseudoobscura*. The males have however, an extraordinary secondary sexual characteristic, unique not only among species of *obscura* and *affinis* groups but also in the genus *Drosophila* as a whole. The new species is named in honor of Cuauhtemoc, the last emperor of the Aztecs and a national hero of Mexico.

Drosophila cuauhtemoci, new species, Felix & Dobzhansky

Male.—Head with arista with 7 branches (3 above, 2 below, and the terminal fork); antennae brown, 3rd. joint darker; front dark brown; middle orbital less than half as long as posterior; one prominent oral bristle; carina broadening below, subtriangular; cheeks brown, their greatest width about 1/8 of greatest diameter of eye; eyes dark red; in living specimens brighter than in *D. pseudoobscura* and *D. azteca*.

Thorax with 8 rows of blackish brown acrostichal hairs; anterior scutellars convergent; mesonotum brownish black, with an indistinct lighter band in the middle; pleurae blackish brown, lighter posteriorly (longitudinal). Legs blackish brown; two small sex combs on the front tarsi (Fig. 1A), comb on the first tarsal joint usually with 4 but sometimes with 5 teeth, comb on the second joint

[1] Research supported by National Science Foundation Grant OIP-75-06738 and El Consejo Nacional de Ciencia y Technologia de Mexico.

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with 3 teeth, the bases of the teeth in lines almost perpendicular to the axis of the tarsal joints; tibiae with preapical bristles; tibiae of the middle legs broadened, with rows of long bristles on either side (Fig. 1B). Wings transparent; costal index average 2.46 (limits 2.27–2.56); 4th vein index 2.00 (1.87–2.15); 4c index 1.03 (0.97–1.08); 5x index 1.94 (1.76–2.23), appears to be greater in dried specimens than in those cleared in glycerine; length 2.45 mm (2.22–2.54).

Abdomen dark brownish black, the last tergite more shiny than the preceeding ones; genital arch (Fig. 2A) dark brown; toe rather short, rounded; clasper comb with 9 dark teeth; aedeagus (Fig. 2B) long and slender, parameres long, acuminate at the end; novasternum large, outwardly flattened. Internal genitalia with testes bright orange, ellipsoidal or banana-shaped, seminal vesicles long, bright orange.

Body length 1.85 mm in dried specimens, 2.65 in specimens cleared in glycerine.

Chromosomes.—No satisfactory spermatogonial metaphase plates have been obtained in preparations of testes of wild-collected males. The few dividing cells that were observed appeared to show chromosomes similar to those of *D. pseudoobscura.*

Geographic Distribution.—The original specimens of *D. cuauhtemoci* males (including the type) were collected on July 29th, 1974, in a mixed pine and oak forest 10 km north of Cuernavaca along Highway 95, between Cuernavaca and the city of Mexico. Other collections were made in the same neighborhood on several occasions in 1974 and 1975. Some individuals were found near Lago Patzcuaro, Michoacan, and in Parque Nacional El Chico, near Pachuca, Hidalgo.

Females: A majority of the flies in the population samples containing males of *D. cuauhtemoci* belonged to the species *D. pseudoobscura* and *D. azteca.* Since females of these species are not easily distinguishable, they were placed individually in bottles with *Drosophila* culture medium. Only one of the undoubtedly numerous females of *D. cuauhtemoci* produced progeny; she was identified as belonging to that species because her sons were unmistakably *D. cuauhtemoci.* Two of her daughters, preserved in alcohol, have been carefully examined. No clear-cut external differences from *D. pseudoobscura* females, except perhaps for brighter red eyes, were found. Internally, the spermathecae are darker and less convex than in *D. pseudoobscura.* The holotype is deposited at the California Academy of Sciences, San Francisco. Paratypes at the Department of Entomology, University of California, Davis, the American Museum, New York City, and the National Museum of Natural History, Washington, D.C.

Ayala and Powell (1972) have shown that sibling species of the *obscura* and *willistoni* groups can be diagnosed by enzyme patterns detected by electrophoresis. Ayala and Tracey (1974) and Ayala and

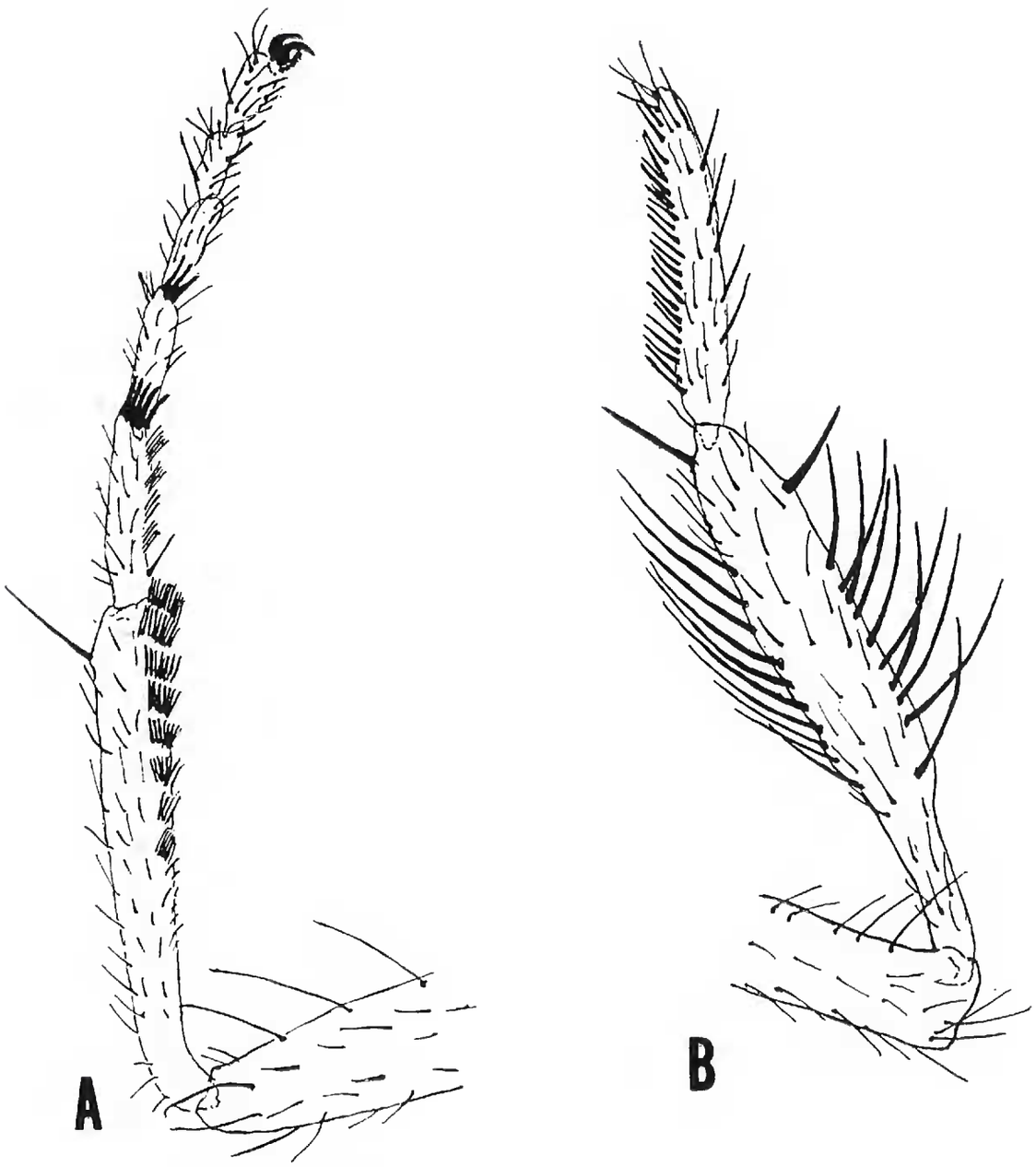

FIG. 1. The tibia and tarsus of the anterior leg (A), and the tibia and the first tarsal joint (B) of *Drosophila cuauhtemoci.*

Dobzhansky (1974) found differences in frequencies of certain allozymes between subspecies of *D. pseudoobscura, D. willistoni,* and *D. equinoxialis.* We have used 8 males of *D. cuauhtemoci* and determined the electrophoretic pattern of 8 enzyme systems for this species. These were run simultaneously with, and compared to, *D. pseudoobscura, D. persimilis* and *D. azteca.*

Three of the eight systems showed clear-cut differences between *D. cuauhtemoci* and *D. pseudoobscura, D. persimilis* and *D. azteca*: (a) Leucine amino peptidase (Lap): If we designate as a standard the most common allele in *D. pseudoobscura* and *D. persimilis* as 1.00, all

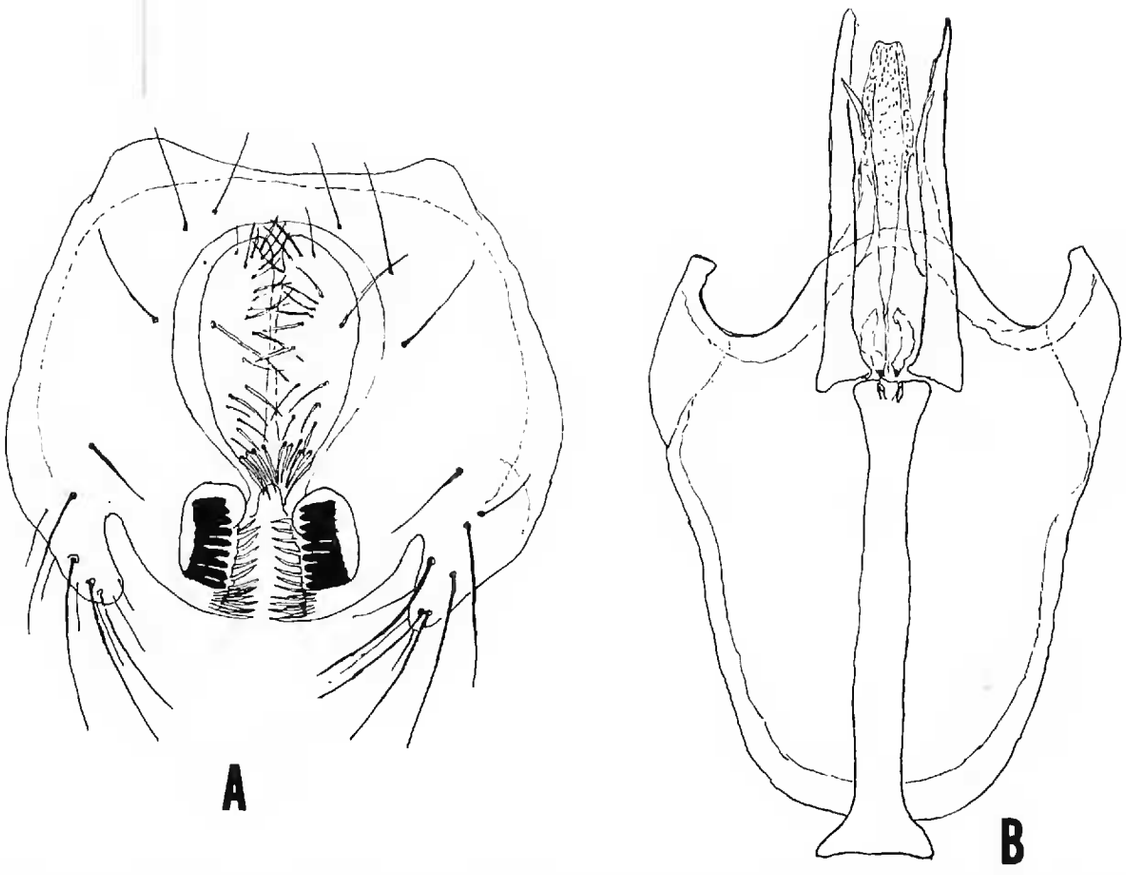

FIG. 2. The genital arch and anal plates (A), and the aedeagus with the adjacent parts (B) of *Drosophila cuauhtemoci.*

8 *D. cuauhtemoci* specimens examined were homozygous for an allele 1.12; that is, it traveled 12% faster in our gel system. (Details of the procedures and systems can be found in Ayala et. al., 1972). *D. azteca* was polymorphic for two alleles, 1.17 and 1.22 relative to the standard. (b) Malic dehydrogenase (Mdh): Again using the *D. pseudoobscura* and *D. persimilis* most common allele as the standard 1.00, all 8 *D. cuauhtemoci* males were homozygous for allele 1.18. *D. azteca* was polymorphic for two alleles at the Mdh locus, 1.16 and 1.36.
(c) Alcohol dehydrogenase (Adh): This enzyme migrated cathodally in our electrophoretic system and the most common allele in *D. pseudoobscura* and *D. persimilis* is designated –1.00 as a standard. All *D. cuauhtemoci* specimens were homozygous for allele –1.23. *D. azteca's* most common allele is –1.40.

The resolution of alkaline phosphatase was not good enough to quantify differences, but *D. cuauhtemoci* did appear to be different from the other *obscura* group species tested. Esterases are very polymorphic in all the *obscura* group species. *D. cuauhtemoci* has alleles in common with *D. pseudoobscura, D. persimilis* and *D. azteca*; however, the sample size was not large enough to determine if the frequencies of alleles are

greatly different in *D. cuauhtemoci* as compared to the other species. For the following enzymes, all four *obscura* group species examined were homozygous for the same electrophoretic allele: α-glycerophosphate dehydrogenase, tetrazolium oxidase, and acid phosphatase.

D. cuauhtemoci is clearly a close relative of *D. pseudoobscura*, the females of the two species being practically indistinguishable by externally visible traits. The males are however, easily identifiable by the tibiae of their middle legs, their sex combs, and their genitalia. Curiously enough, some of their characteristics are intermediate between the *obscura* and the *affinis* groups. (*D. azteca* is a member of the latter group.) The distal sex comb is smaller in *D. cuauhtemoci* than in any species of the *obscura* group, while *affinis* species usually have a single tooth in place of the distal sex comb. The testes of *D. cuauhtemoci* are more elongate than in *D. pseudoobscura* and its relatives, while it is spiral in *affinis* group species. And finally the electrophoretic mobility of leucine amino peptidase, malic dehydrogenase, and alcohol dehydrogenase of *D. cuauhtemoci* is intermediate between those of *D. pseudoobscura* and *D. persimilis* on the one hand and *D. azteca* on the other. *D. cuauhtemoci* could almost be considered a species close to the common ancestry of the *obscura* and *affinis* species group, yet its unique and specialized secondary sexual character is not found even as a rudiment in these species groups.

ACKNOWLEDGMENTS

It is a pleasure to thank Dr. Alfonso L. de Garay, Director of the Departamento de Radiobiologia y Genetica, INEN, Mexico, for his support of this research program on the study of Mexican *Drosophila*. We wish to thank also the authorities of INEN for the laboratory and collecting facilities used in this study.

LITERATURE CITED

AYALA, F. J., AND TH. DOBZHANSKY. 1974. A new subspecies of *Drosophila pseudoobscura* (Diptera: Drosophilidae). Pan-Pacific Entomol., 50: 211–219.

AYALA, F. J., AND J. R. POWELL. 1972. Allozymes as diagnostic characters of sibling species of *Drosophila*. Proc. Nat. Acad. Sci. USA, 69: 1094–1096.

AYALA, F. J., J. R. POWELL, M. L. TRACEY, C. A. MOURAO, AND S. PEREZ-SALAS. 1972. Genic variation in natural populations of *Drosophila willistoni*. Genetics, 70: 113–139.

AYALA, F. J., AND M. L. TRACEY. 1974. Genetic differentiation and reproductive isolation between two subspecies. J. Heredity, 64: 120–124.